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The Panamanian Aotus - Plasmodium falciparum model was used to evaluate potential antimalaria drugs. Neither protriptylene nor tetrandrine, each administered with chloroquine, was effective in reversing chloroquine, resistance of Vietnam Smith/RE infections. Promethazine, administered twice daily, with chloroquine was no more effective in reversing chloroquine-resistance than when administered once daily. The absence of antimalarial activity of chlorpromazine and prochlorperazine confirmed previous studies in which in vivo reversal of chloroquine resistance was attributed to each of these drugs plus chloroquine. A bisquinoline, WR 268668, was inactive against Smith/RE infections, possibly due to lack of absorption. A water soluble form of the drug is scheduled to be evaluated.

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INTRODUCTION

The essence of the problem addressed in this report is to evaluate the potential antimalarial activity of drugs in the preclinical model of Aotus lemurinus lemurinus (Panamian night monkey) experimentally infected with Plasmodium falciparum. Such studies with this model were initiated in 1976 at Gorgas Memorial Laboratory, Panama and supported, in part, by the U.S. Army Medical Research and Development Command. Due to the drug resistance exhibited by the highly pathogenic P. falciparum parasites in Asia, Africa, and Latin America, it is essential that new drugs be evaluated in the preclinial Aotus model for their potential usefulness against human infections.

Initially, antimalarial drug studies used the Colombian Aotus as the experimental host (1,2). In the mid 1970's, embargoes imposed by South American countries on the exportation of monkeys seriously restricted the use of Aotus for biomedical research in the United States. Panamanian Aotus were available at Gorgas Memorial Laboratory, Panama, and the projected transferred here in 1976. Diverse avenues of research have been pursued in attempts to identify effective new antimalarial drugs. Three strains of P. falciparum, Vietnam Smith, Uganda Palo Alto, and Vietnam Oak Knoll, had been adapted to Panamanian Aotus. These strains exhibit diverse susceptibility and/or resistance to standard antimalarial agents. The course of untreated infections in Panamanian Aotus has been characterized and compared with that in Aotus of Colombia (3). Overall, the virulence of these strains was less in Panamanian than in Colombian owl monkeys, as indicated by lower mortality rates of Panamanian monkeys during the first 30 days of patency. Maximum parasitemias of the Vietnam Smith and Uganda Palo Alto strains were, however, significantly higher during the first 15 days of patency in Panamanian than in Colombian owl monkeys. These quantitative differences in infection parameters between Panamanian and Colombian owl monkeys have not invalidated the use of the former for the evaluation of new antimalarial drugs.

Numerous candidate antimalarial drugs of diverse chemical classes have been evaluated against trophozoite-induced infections of one or more P. falciparum strains during the course of these contracts. In seeking alternatives to primaquine, two 8-aminoquinolines proved to be active against the blood stages of P. falciparum (4,5). Desferrioxamine, an iron-specific chelating agent, was shown to suppress parasitemias of the virulent Uganda Palo Alto strain of P. falciparum (6). The in vitro activity of two halogenated histidine analogs was not confirmed by evaluation against P. falciparum infections in owl monkeys (7).

Chloroquine-resistance of P. falciparum represents the greatest challenge in developing effective antimalarial drugs. Reversal of chloroquine-resistance in P. falciparum, in vitro, was achieved by the co-administration of verapamil (a calcium channel blocker) plus

chloroquine (8). Other in vitro studies have shown that there is a significantly greater efflux of chloroquine from erythrocytes containing falciparum parasites resistant to chloroquine than from red cells parasitized by chloroquine-sensitive falciparum malaria (9). Calcium channel blockers appear to prevent this active efflux of chloroquine, thus allowing the drug to accumulate to parasiticidal levels.

Based upon the success of in vitro reversal of chloroquine-resistance, trials were initiated to determine if resistance could be reversed in Aotus infected with the chloroquine-resistant Vietnam Smith strain of P. falciparum. Six calcium channel blockers, or similarly acting drugs, were co-administered with chloroquine in diverse regimens. The desideratum of chloroquine-resistance reversal was administration of a single course of treatment, with parasite clearance and infection cure. Suppression of parasitemia was obtained during an initial course of treatment, but parasite clearance and cure occurred in some instances only after re-treatment. Such infection parameters were similar to those in monkeys with self-limited infections and cure could be attributed to acquired immunity.

Limited trials with desipramine, Norpramin, a tricyclic psychotropic drug, demonstrated the feasibility of reversing chloroquine-resistance in vivo (10). Parasite clearance was obtained, but the infection was not cured.

Subsequently, in vivo reversal of chloroquine resistance was obtained with combinations of chloroquine plus chlorpromazine or prochlorperazine. Such reversal was exhibited by rapid suppression and clearance of parasitemia, resulting in infection cure without retreatment.

Evaluation of two oil-soluble derivatives of artemisinin, artemether and arteether, demonstrates that both possess similar activity to cure infections of a multi-drug resistant P. falciparum strain in Aotus.

Both the purpose and methods of approach of the present work remains essentially unchanged since 1976, viz to ascertain the antimalarial activity of drugs against P. falciparum infections in Aotus. The method of approach may vary on an ad hoc basis, such as administering a combination of drugs.

Before ending this section, I believe a summary of events that occurred some months prior to the initiation of this contract is in order. As referred to previously, the antimalarial drug project began in 1976 at Gorgas Memorial Laboratory with U.S.A.M.R.D.C. funds awarded to Gorgas Memorial Institute (G.M.I.), Bethesda, M.D. On 15 December 1990, Gorgas Memorial Laboratory ceased to function under G.M.I. and the physical facilities became the responsibility of the Panamanian Ministry of Health. The Minister of Health approved

a request that the Aotus colony and the program continue at Gorgas Memorial Laboratory. A new research contract proposal was prepared indicating that the administration of funds was to be by PROMED TRADING, S.A., a commercial firm located in Panama. The time required for contract proposal preparation, approval and funding was estimated to be approximately five months. I must express my profound appreciation to those individuals at the Division of Experimental Therapeutics, WRAIR, and at the USAMRDC, Ft. Detrick, for the unceasing and successful efforts to obtain interim funding until the contract was awarded and funded. Without their diligence, the program would have terminated.

BODY

I. Experimental Methods

The general intent of this project is to evaluate the potential antimalarial activity of drugs, or combination thereof, in the pre-clinical model of Aotus experimentally infected with P. falciparum (or P. vivax). Specifically, the vertebrate host is Aotus lemurinus lemurinus, the Panamanian night monkey. These animls are either feral, laboratory adapted or laboratory born. No naturally acquired, human plasmodium infection has been reported in Aotus. The Vietnam Smith/RE strain of P. falciparum was adapted to Aotus of Colombian origin in 1971(1) and in Panamanian Aotus in 1976.(3) The course of untreated infections, essential for comparison with treated infections, has been documented in Panamanian Aotus (3). This plasmodium strain is resistant to maximally tolerated doses of chloroquine, pyrimethamine, and quinine (2).

To initiate an experiment, infected blood (with 2.5% sodium citrate as the anticoagulant) from an untreated Aotus was diluted appropriately in chilled saline (0.85%), such that each milliliter contained 5,000,000 parasites. This amount was inoculated into the saphenous vein of experimental and control monkeys.

Blood films, prepared and examined daily beginning on the first post-inoculation day, were stained with Giemsa. Parasitemias were evaluated as follows: negative, if no parasites were detected on a thick blood film after examination for at least 5 minutes; <10 parasites per cmm, if positive only on the thick blood film; parasite enumeration was by the Earle-Perez method and reported as the number of parasites per cmm. (11)

Blood films from untreated Aotus, serving as passage and/or control subjects, were prepared and examined daily during the primary patent period, and daily thereafter for at least three consecutive days after parasites could last be detected on thick blood films. When parasitemia had cleared, films were made and examined twice weekly until a total of 100 negative days had been recorded. If a recrudescence occurred, blood films were obtained again on a daily basis.

Parasitemias were evaluated daily during the treatment period and until blood films were negative for at least seven consecutive days. The frequency of smearing was then reduced to two time per week (Monday and Thursdays or Tuesdays and Fridays). If no recrudescences occurred during a 100 day examination period, the infection was considered to have been cured.

Drug doses were calculated as mg base per kg of body weight. Stock solutions of water soluble compounds, at appropriate concentrations, were prepared with distilled water and stored at 8°C for the treatment period. If a compound was water insoluble, a

suspension of the requisite amount of drug was prepared daily with 0.3% methylcellulose (in distilled water).

Oral administration of drugs was effected by gastric intubation with a 14 French catheter. The total amount of fluid administered, drug solution or suspension, and rinse was 14 ml.

II. Results

A. WR 035941AB (BN: BL18130), protriptylene
WR 149557AB (BN: BM06813), tetrandrine

This study was a continuation of trials to reverse chloroquine-resistance in vivo by the concomitant administration of the experimental drug plus chloroquine. Prior to testing in infected monkeys, the drug combinations were evaluated for toxicity in malaria-cured monkeys, as determined by body-weight changes shown in Table 1. There was no evidence of drug intolerance, such as vomiting, anorexia, or loss of motor function. Within one month after the drugs had been administered, the monkey body weights were essentially equal to the pre-experimental levels. The drug combinations were considered to be non-toxic and experiments initiated.

The detailed response of Vietnam Smith/RE parasites to WR 035941 (protriptylene) plus WR 1544 (chloroquine) is given in Table 2 and summarized in Table 3. When the drug combination was administered once daily for 7 days during the ascending phase of the parasitemia, clearance occurred in 3 of 3 Aotus, and one infection was cured. Retreatment with twice the dose of protriptylene and the daily maximum tolerated dose (20.0 mg/kg) of chloroquine cleared parasitemia in 2 of 2 animals and cured the infection in one monkey.

B. WR 002158AJ (BN: BL 50610), promethazine

Based upon anecdotal observations in Africa, the concomitant administration of promethazine (Phenergan), an antihistamine, plus chloroquine, at least ameliorates P. falciparum infections. In a previous trial using the Aotus - P. falciparum model, promethazine (10.0 mg/kg, once daily for 7 days) plus chloroquine (20.0 mg/kg, once daily for 7 days) only cleared the parasitemia in 1 of 2 Aotus. A similar drug regimen, but with 20.0 mg/kg of promethazine cleared parasitemia in 1 of 2 Aotus, but the infection was not cured. A second experiment, evaluated this drug combination as previously, except that during the primary treatment regimen, promethazine was administered twice daily, at 8:00 AM and 4:00 PM.

The detailed antimalarial activity of promethazine plus chloroquine is shown in Table 6 and summarized in Table 7. A 10.0 mg/kg dose of promethazine, twice daily, for seven days plus a daily 20.0 mg/kg dose of chloroquine, cleared parasitemia (without cure) in 1 of 2 Aotus. Parasitemia in each of 2 Aotus was cleared,

again without cure, when the dose of promethazine was increased to 20.0 mg/kg, twice daily administered with chloroquine. The results of the primary treatments were essentially the same as in the first pilot evaluation. Accordingly, during retreatments, promethazine was administered once daily.

Data in Table 7 indicate that retreatments cleared parasitemia in 6 of 6 Aotus, but the infection was cured in only one animal after a total three treatments.

C. WR 002173AL(BN: BK 20886), chlorpromazine
WR 006379AF(BN: BM 01907), prochlorperazine

In studies previously reported, each of these phenothiazines administered at a dose of 20.0 mg/kg once daily for 7 days with chloroquine (20.0 mg/kg x 7 days) during the ascending phase of the parasitemia reversed chloroquine resistance in vivo as shown by parasite clearance and infection cure. An important lacuna in those experiments was that the antimalarial activity, or lack thereof, for each of the drugs alone was not determined. Two experiments were initiated to obtain such essential data.

Each of two Aotus, infected with the chloroquine-resistant Vietnam Smith/RE strain of P. falciparum, received chlorpromazine, 20.0 mg/kg, once daily for 7 days. The data in Tables 8 and 9 indicate that this dose had no effect upon the parasitemia, nor did administration of 2 x the original dose as both animals succumbed to a fulminating infection. The resistance to chloroquine of this strain was re-confirmed by the lack of parasite response to the maximum daily tolerated dose of chloroquine - 20.0 mg/kg.

Prochlorperazine alone (20.0 mg/kg, once daily x 7 days) had no effect upon the ascending phase of Vietnam Smith/RE parasitemia (Tables 10 and 11). Retreatment with 40.0 mg/kg of the drug had no activity in one Aotus and suppressed parasitemia in one animal which died of malaria. Again, chloroquine alone possessed no antimalarial activity.

D. WR 268668AC (BN:BM 10586)

This bisquinoline was demonstrated in other laboratories to possess antimalarial activity against chloroquine-resistant strains of P. falciparum, both in vitro and in the rodent malaria model. This water insoluble drug, administered as a suspension in 0.3% methyl cellulose, had no activity or suppressive activity at doses of 1.0, 4.0, and 16.0 mg/kg, once daily, for three days, against Vietnam Smith/RE primary parasitemias. (Tables 13 and 14). Re-treatment with a dose of 4.0 mg/kg had no effect upon parasitemia in 2 of 2 Aotus, a dose of 16.0 mg/kg cleared parasitemia (without cure) in 1 of 2 Aotus, and a dose of 64.0 mg/kg cleared parasitemia in 3 of 5 monkeys, curing the infection in one subject (Table 15).

III. Discussion

The desideratum to demonstrate in vivo reversal of chloroquine-resistance is to administer the experimental drug plus chloroquine, in a prescribed regimen, during the primary ascending phase of the parasitemia. The parasitemia must be cleared and the infection cured, as indicated by the absence of recrudescence for at least 100 days after blood films are parasite negative. Obviously, neither the experimental drug alone, nor chloroquine should possess any antimalarial activity in the test model. Results of retreatment after the initial peak infection and of recrudescences are difficult to interpret due to acquired immunity leading to self-cure of untreated infections. In the case of Panamanian Aotus infected with the Vietnam Smith strain, 72% of the infections are self-limited (3).

Protriptylene (WR 035941AB) plus chloroquine (WR 1544BM) cured 1 of 3 infections of the chloroquine-resistant Vietnam Smith/RE strain, while tetrandrine (WR 149557AB) plus chloroquine cured 0 of 3 such infections when administered during the primary parasitemia. Neither experimental drug has the capacity to reverse chloroquine-resistance of a human plasmodium to an extent that would warrant further testing.

A second trial using promethazine (WR 002158) plus chloroquine differed from the initial trial in that promethazine was administered twice daily, instead of once a day. Comparison of results between the two trials showed no essential difference when promethazine was administered twice daily. Primary treatment did not cure infections. Recent pharmacokinetic studies with promethazine and chloroquine indicate that to possibly achieve chloroquine resistance reversal, a loading dose of chloroquine should be administered at 8:00 AM, followed by two doses of promethazine, each at 4 hr intervals. Such a trial using this regimen is projected to be initiated shortly in the Aotus model.

The absence of antimalarial activity of chlorpromazine (WR 002173) and prochlorperazine (WR 006379) against Vietnam Smith/RE infections confirms results of prior studies which showed that when each of these drugs is administered with chloroquine, parasitemias were cleared and infections cured. This was a clear demonstration of in vivo reversal of chloroquine-resistance.

The inactivity of WR 268668 was disappointing in view of its efficacy in vitro and in the rodent malaria model. Since the drug was insoluble in water and administered orally as a suspension, it is suggested that little or no drug was absorbed through the Aotus intestine. To test this hypothesis, a water soluble form of the drug has been prepared and will be evaluated by oral and parenteral (intramuscular) administration against Vietnam Smith/RE infected Aotus.

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TABLE 1

TOXICITY EVALUATION OF WR 035941AB(BL 18130), PROTRIPTYLENE, AND WR 149557AB(BM 06813) TETRANDRINE, BOTH IN COMBINATION WITH WR 154480M(AR 20613), CHLOROQUINE

Monkey No.	Drug mg/kg	Body Weight (gms) Days Post Treatment				
		-7	7	15	22	29
12301	19.0a	723	678	697	698	746
12347	19.0a	738	730	745	761	760
12306	15.0b	854	809	835	830	825
12313	15.0b	680	642	648	660	668

a. WR 035941AB, oral, once daily x 7 days

b. WR 149557AB, oral, once daily x 7 days

Each monkey also received chloroquine, 20.0 mg/kg, oral, once daily x 7 days

TABLE 2

DETAILED ACTIVITY OF WR 035941AB(BN:BL 18130) PLUS WR 1544BM (BN:AR 20613) AGAINST INFECTIONS OF THE VIETNAM SMITH/RE STRAIN OF PLASMODIUM FALCIPARUM

Aotus No.	Daily Dose Mg/Kg	Parasitemia per cmm x 10^3										
		Day Pre-Rx	Day of Treatment							Day Post Treatment		
			1	2	3	4	5	6	7	1	2	3
87032	19.0a 20.0b	4	19	16	5	0.2	< 0.01	<0.01	0	0	0	0
89034	19.0a 20.0b	39	55	237	76	0.3	< 0.01	< 0.01	0	0	0	0
89067	19.0a 20.0b	20	9	4	1	<0.01	< 0.01	<0.01	0	0	0	0
87032r	38.0a 20.0b	1	11	4	< 0.01	0	0	0	0	0	0	0
89034r	38.0a 20.0b	< 0.01	0.5	4	14	1	< 0.01	0	0	0	0	0

a WR 035914 AB
b WR 1544BM

TABLE 3

SUMMARY OF THE ACTIVITY OF WR 035941AB(BL 18130), PROTRIPTYLENE, PLUS WR 1544BM(AR 20613), CHLOROQUINE, AGAINST INFECTIONS OF THE VIETNAM SMITH/RE STRAIN OF PLASMODIUM FALCIPARUM

Monkey No.	Dose x 7 Mg/Kg	Response of Parasitemia to Rx			Days from Initial Rx to Parasite Clearance	Days from Final Rx To Recru-descence	Notes
		None	Suppressed	Cleared			
87032	19.0a 20.0b			+	7	25	Re-Rx
89034	19.0a 20.0b			+	7	19	Re-Rx
89067	19.0a 20.0b			+	7	n.a.	Cured
87032r	38.0a 20.0b			+	4	n.a.	Cured
89034r	38.0a 20.0b			+	6	20	

a WR 035914AB

b WR 1544BM

TABLE 4

DETAILED ACTIVITY OF WR 149557AB(BN: 06813) PLUS WR 1544BM (BN:AR 20613) AGAINST INFECTIONS OF THE VIETNAM SMITH/RE STRAIN OF PLASMODIUM FALCIPARUM

Aotus No.	Daily Dose Mg/Kg	Parasitemia per cmm x 10^3									
		Day Pre-Rx	Day of Treatment							Day Post Treatment		
			1	2	3	4	5	6	7	1	2	3
87025	15.0a 20.0b	6	95	331	181	150	42	23	<0.01	<0.01	<0.01	<0.01
87055	15.0a 20.0b	15	54	314	104	32	3	<0.01	<0.01	0	0	0
88016	15.0a 20.0b	22	39	147	26	2	0.3	<0.01	<0.01	0	0	0
87025r	30.0a 20.0b	<0.01	<0.01	<0.01	<0.01	0	0	0	0	0	0	0
88016r	30.0a 20.0b	3	4	0.9	0.2	<0.01	<0.01	0	0	0	0	0

a WR 149557AB

b WR 1544BM

TABLE 5

SUMMARY OF THE ACTIVITY OF WR 149557AB(BM 06813), TETRANDRINE, PLUS WR 1544BM(AR 20613), CHLOROQUINE, AGAINST INFECTIONS OF THE VIETNAM SMITH R/E STRAIN OF PLASMODIUM FALCIPARUM

Monkey No.	Dose x 7 Mg/Kg	Response of Parasitemia to Rx			Days from Initial Rx to Parasite Clearance	Days from Final Rx To Recru-descence	Notes
		None	Suppressed	Cleared			
87025	15.0a 20.0b			+	16	23	Re-Rx
87055	15.0a 20.0b			+	8	33	
88016	15.0a 20.0b			+	8	13	Re-Rx, higher dose
88016r	30.0a 20.0b			+	6	23	
87025r	30.0a 20.0b			+	4	n.a.	Cured

a WR 149557AB

b WR 1544BM

TABLE 6

DETAILED ACTIVITY OF WR 2158AJ(BN: BL 50610), PROMETHAZINE, PLUS WR 1544BM(BN: AR20613), CHLOROQUINE, AGAINST INFECTIONS OF THE VIETNAM SMITH/RE STRAIN OF PLASMODIUM FALCIPARUM

Aotus No.	Daily Dose Mg/Kg	Parasitemia per cmm x 10^3										
		Day Pre-Rx	Day of Treatment							Day Post Treatment		
			1	2	3	4	5	6	7	1	2	3
84055	10.0a 20.0b	2	10	79	40	52	31	1	<0.01	<0.01	<0.01	1
89022	10.0a 20.0b	30	42	251	41	1	<0.01	<0.01	0	0	0	0
86040	20.0a 20.0b	0.7	4	3	3	0.2	<0.01	<0.01	0	0	0	0
89014	20.0a 20.0b	27	55	109	51	6	0.2	<0.01	0	0	0	0
84055r	20.0c 20.0b	133	24	37	74	6	3	0.9	<0.01	<0.01	<0.01	0
89022r	20.0c 20.0b	<0.01	1	2	0.5	0.6	0.1	<0.01	0	0	0	0

TABLE 6 (CONT'D.)

DETAILED ACTIVITY OF WR 2158AJ(BN: BL 50610), PROMETHAZINE, PLUS WR 1544BM(BN: AR20613), CHLOROQUINE, AGAINST INFECTIONS OF THE VIETNAM SMITH/RE STRAIN OF PLASMODIUM FALCIPARUM

Aotus No.	Daily Dose Mg/Kg	Parasitemia per cmm x 10^3									
		Day Pre-Rx	Day of Treatment							Day Post Treatment		
			1	2	3	4	5	6	7	1	2	3
89014r	40.0c 20.0b	<0.01	3	5	0.8	3	0.4	<0.01	<0.01	0	0	0
86040r	40.0c 20.0b	2	108	34	71	9	26	4	4	0.2	<0.01	<0.01
89022rr	40.0c 20.0b	<0.01	0.1	0.3	<0.01	0	0	0	0	0	0	0
89014rr	80.0c 200b	<0.01	<0.01	<0.01	<0.01	0	0	0	0	0	0	0

a WR 2158AJ, twice daily

b WR 1544BM, once daily

c WR 2158AJ, once daily

TABLE 7

SUMMARY OF THE ACTIVITY OF WR 2158AJ(BL 50610), PROMETHAZINE, PLUS WR 1544BM(AR 20613), CHLOROQUINE, AGAINST INFECTIONS OF THE VIETNAM SMITH/RE STRAIN OF PLASMODIUM FALCIPARUM

Monkey No.	Dose x 7 Mg/Kg	Response of Parasitemia to Rx			Days from Initial Rx to Parasite Clearance	Days from Final Rx To Recru-descence	Notes
		None	Suppressed	Cleared			
84055	10.0a 20.0b		+		n.a.	n.a.	Re-Rx, higher dose
89022	10.0a 20.0b			+	7	7	Re-Rx, higher dose
86040	20.0a 20.0b			+	7	17	Re-Rx, higher dose
89014	20.0a 20.0b			+	7	8	Re-Rx, higher dose
84055r	20.0c 20.0b			+	10	n.a.	Cured
89022r	20.0c 20.0b			+	7	9	Re-Rx, higher dose
89014r	40.0c 20.0b			+	8	8	Re-Rx, higher dose
86040r	40.0c 20.0b			+	15	63	
89022rr	40.0c 20.0b			+	4	28	
89014rr	80.0c 20.0b			+	5	n.a.	Cured

a WR 2158AJ 8:00 AM & 4:00 PM
b WR 1544BM once daily
c WR 2158AJ once daily

TABLE 8

DETAILED ACTIVITY OF WR 002173AL(BN:BK 20886), CHLORPROMAZINE AGAINST INFECTIONS OF THE VIETNAM SMITH/RE STRAIN OF PLASMODIUM FALCIPARUM

Aotus No.	Daily Dose Mg/Kg	Parasitemia per cmm x 10^3										
		Day Pre-Rx	Day of Treatment							Day Post Treatment		
			1	2	3	4	5	6	7	1	2	3
12635	20.0a	8	32	89	351	542	222	641	1183	Re-Rx, higher dose		
12636	20.0a	0.8	1	30	95	114	191	517	316	Re-Rx, higher dose		
12635r	40.0a	1183	823	511	DIED, malaria							
12636r	40.0a	316	1010	956	DIED, malaria							
12631	20.0b	13	95	97	242	189	240	265	296	345	567	345

a WR 002173AL

b WR 1544BM, chloroquine

TABLE 9

SUMMARY OF THE ACTIVITY OF WR 002173AL(BK 20886), CHLORPROMAZINE AGAINST INFECTIONS OF THE VIETNAM SMITH R/E STRAIN OF PLASMODIUM FALCIPARUM

Monkey No.	Dose x 7 Mg/Kg	Response of Parasitemia to Rx			Days from Initial Rx to Parasite Clearance	Days from Final Rx To Recru-descence	Notes
		None	Suppressed	Cleared			
12635	20.0a	+			n.a.	n.a.	Re-Rx, higher dose
12636	20.0a	+			n.a.	n.a.	Re-Rx, higher dose
12635r	40.0a	+			n.a.	n.a.	Died Day 3 of Rx
12636r	40.0a	+			n.a.	n.a.	Died Day 3 of Rx
12631	20.0b	+			n.a.	n.a.	

a. WR 002173AL
b. Chloroquine, WR 1544BM

TABLE 10

DETAILED ACTIVITY OF WR 006379AF(BN: BK20886), PROCHLORPERAZINE AGAINST INFECTIONS OF THE VIETNAM SMITH/RE STRAIN OF PLASMODIUM FALCIPARUM

Aotus No.	Daily Dose Mg/Kg	Parasitemia per cmm x 10^3									
		Day Pre-Rx	Day of Treatment							Day Post Treatment		
			1	2	3	4	5	6	7	1	2	3
12637	20.0a	2	8	43	177	198	490	296	259	Re-Rx, higher dose		
12638	20.0a	7	24	57	228	388	394	290	333	Re-Rx, higher dose		
12637r	40.0a	259	265	253	83	296	195	439	36	Rx, new drug		
12638r	40.0a	333	339	220	132	DIED, malaria						
12631	20.0b	13	95	97	242	189	240	265	296	345	567	345

a WR 006379AF

b WR 1544BM, chloroquine

TABLE 11

SUMMARY OF THE ACTIVITY OF WR 006379AF(BM 01907), PROCHLORPERAZINE, AGAINST INFECTIONS OF THE VIETNAM SMITH R/E STRAIN OF PLASMODIUM FALCIPARUM

Monkey No.	Dose x 7 Mg/Kg	Response of Parasitemia to Rx			Days from Initial Rx to Parasite Clearance	Days from Final Rx To Recru-descence	Notes
		None	Suppressed	Cleared			
12637	20.0a	+			n.a.	n.a.	Re-Rx, higher dose
12638	20.0a	+			n.a.	n.a.	Re-Rx, higher dose
12637r	40.0a	+	+		n.a.	n.a.	Rx, new drug
12638r	40.0a				n.a.	n.a.	Died Day 4 of Rx
12631	20.0b	+			n.a.	n.a.	

a. WR 006379AF
b. Chloroquine, WR 1544BM

TABLE 12

SUMMARY OF IN VIVO TRIALS TO REVERSE CHLOROQUINE RESISTANCE OF PLASMODIUM FALCIPARUM INFECTIONS

MALARIA STRAIN	DOSE mg/kg TOTAL	DOSE mg/kg DAILY	PRIMARY TREATMENTS CLEARED	PRIMARY TREATMENTS CURED	REPEAT TREATMENTS CLEARED	REPEAT TREATMENTS CURED	TOTAL TREATMENTS CLEARED	TOTAL TREATMENTS CURED
Smith/RE								
	WR 035941AB(a) plus WR 1544BM(b)							
	133.0a 140.0b	19.0a 20.0b	3/3	1/3			3/3	1/3
	266.0a 140.0b	38.0a 20.0b			2/2	1/2	2/2	1/2
	WR 149557AB(c) plus WR 1544BM(b)							
	105.c 140.0b	15.0c 20.0b	3/3	0/3			3/3	0/3
	210.0c 140.0b	30.0c 20.0b			2/2	1/2	2/2	1/2
	WR 2158AJ(d) plus WR 1544BM(b)							
	140.0d 140.0b	20.0d 20.0b	1/2	0/2	2/2	1/2	3/4	1/4
	280.0d 140.0b	40.0d 20.0b	2/2	0/2	3/3	0/3	5/5	0/5
	560.0d 140.0b	80.0d 20.0b			1/1	1/1	1/1	1/1

TABLE 13

DETAILED ACTIVITY OF WR 268668AC(BN: BM10586) AGAINST INFECTIONS OF THE VIETNAM SMITH/RE STRAIN OF PLASMODIUM FALCIPARUM

Aotus No.	Daily Dose Mg/Kg	Parasitemia per cmm x 10^3										
		Day Pre-Rx	Day of Treatment			Day Post Treatment						
			1	2	3	1	2	3	4	5	6	7
12632	1.0	10	35	222	517	690	Re-Rx, higher dose					
12626	1.0	0.1	1	3	117	142	444	Re-Rx, higher dose				
12627	4.0	11	37	139	168	120	185	191	345	Re-Rx, higher dose		
12628	4.0	16	111	172	427	738	Re-Rx, higher dose					
12632r	4.0	690	1158	869	542	464	517	Died malaria				
12626r	4.0	444	553	882	382	394	370	Re-Rx, higher dose				
12629	16.0	2	12	48	131	71	25	23	26	Re-Rx, higher dose		
12633	16.0	9	39	129	314	211	Re-Rx, higher dose					
12627r	16.0	345	494	326	222	44	1	0.6	0	0	0	0
12628r	16.0	738	739	419	493	247	212	167	69	296	Re-Rx, higher dose	
12629r	64.0	26	32	32	1	<0.01	<0.01	<0.01	0	0	<0.01	0
12633r	64.0	211	288	283	26	3	0.4	<0.01	<0.01	<0.01	<0.01	<0.01
12626rr	64.0	370	222	134	34	12	<0.01	<0.01	0	0	0	0
12628rr	64.0	296	63	33	30	1	<0.01	0	0	0	0	0
12637rr	64.0	439	36	36	19	Died, malaria						

TABLE 14

SUMMARY OF THE ACTIVITY OF WR 26866AC(BM 10586)
AGAINST INFECTIONS OF THE VIETNAM SMITH R/E STRAIN
OF PLASMODIUM FALCIPARUM

Monkey No.	Daily Dose x 3 Mg/Kg	Response of Parasitemia to Rx			Days from Initial Rx to Parasite Clearance	Days from Final Rx To Recru-descence	Notes
		None	Suppressed	Cleared			
12626	1.0	+			n.a.	n.a.	Re-Rx, higher dose
12632	1.0	+			n.a.	n.a.	Re-Rx, higher dose
12627	4.0		+		n.a.	n.a.	Re-Rx, higher dose
12628	4.0	+			n.a.	n.a.	Re-Rx, higher dose
12626r	4.0	+			n.a.	n.a.	Re-Rx, higher dose
12632r	4.0	+			n.a.	n.a.	Died Day 2 Post-Rx, malaria
12629	16.0		+		n.a.	n.a.	Re-Rx, higher dose
12633	16.0		+		n.a.	n.a.	Re-Rx, higher dose
12627r	16.0			+	7	29	*
12628r	16.0		+		n.a.	n.a.	Re-Rx, higher dose
12629r	64.0			+	7	24	*
12633r	64.0		+		n.a.	n.a.	*
12626rr	64.0			+	7	n.a.	Cured
12628rr	64.0			+	6	27	*
12637rr	64.0		+		n.a.	n.a.	Died Day 1 Post Rx, malaria

* Cured with mefloquine

TABLE 15

SUMMARY OF THE ACTIVITY OF WR 268668AC
(BN: BM10586) AGAINST *PLASMODIUM FALCIPARUM* INFECTIONS

MALARIA	DOSE mg/kg		PRIMARY TREATMENTS		REPEAT TREATMENTS		TOTAL TREATMENTS	
STRAIN	TOTAL	DAILY	CLEARED	CURED	CLEARED	CURED	CLEARED	CURED
Smith/RE	3.0	1.0	0/2	0/2			0/2	0/2
	12.0	4.0	0/2	0/2	0/2	0/2	0/4	0/4
	48.0	16.0	0/2	0/2	1/2	0/2	1/4	0/4
	192.0	64.0			3/5	1/4	3/5	1/4